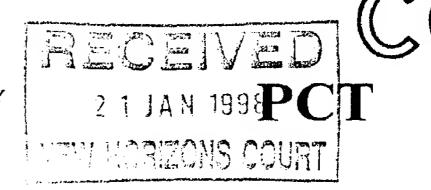
PATENT COOPERATION TREATY

From the

INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:

GIDDINGS, Peter J. SMITHKLINE BEECHAM Corporate Intellectual Property Two New Horizons Court Brentford Middlesex TW8 9EP GRANDE BRETAGNE



NOTIFICATION OF TRANSMITTAL OF INTERNATIONAL PRELIMINARY **EXAMINATION REPORT**

3718

(PCT Rule 71.1)

IMPORTANT NOTIFICATION

Date of mailing (day|month|year)

Applicant's or agent's file reference

AMD/C70237

International application No.

International filing date (day/month/year)

Priority date (day/month/year)

PCT/EP 96/04807

04/11/1996

03/11/1995

Applicant

SMITHKLINE BEECHAM P.L.C. et al.

- 1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.
- 2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.
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4. REMINDER

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

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INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International
AMD/C70237	OR FORTHER ACTION	Preliminary Examination Report (Form PCT/IPEA/416)
International application No.	International filing date (day/n	nonth/year) Priority date (day/month/year)
PCT/EP 96/ 04807	04/11/1996	03/11/1995
International Patent Classification (IPC) or	national classification and IPC	
	C12N15/82	
Applicant		
SMITHKLINE BEECHAM P.L.C	. et al.	
Authority and is transmitted to the	applicant according to Article 3	
2. This REPORT consists of a total	l of sheets, including	this cover sheet.
been amended and are the ba (see Rule 70.16 and Section 6	usis for this report and/or sheets of the Administrative Instruc	of the description, claims and/or drawings which have containing rectifications made before this Authority ctions under the PCT).
These annexes consists of a total of	of sheets.	
 This report contains indications an 	nd corresponding pages relating to	o the following items:
I X Basis of the report		
II Priority		
III Non-establishment of o	opinion with regard to novelty, in	ventive step and industrial applicability
IV Lack of unity of invent	tion	
	der Article 35(2) with regard to sons supporting such statement	novelty, inventive step or industrial applicability;
VI Certain documents cite	d	
VII Certain defects in the in	nternational application	
VIII Certain observations of	n the international application	
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Date of submission of the demand	Date	of completion of this report
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D-80298 Munich Tel. (+49-89) 2399-0, Tx: 5236	656 epmu d	5. 16g13
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INTERNATIONAL PRELIMINARY EXAMINATION REPORT

I. Basis of the report	
	eplacement sheets which have been furnished to the receiving and are referred to in this report as "originally filed" and are ain amendments.):
[] the international application as originall	y filed.
[x] the description, pages 1-50	, as originally filed,
	, filed with the demand,
	, filed with the letter of,
pages	, filed with the letter of,
[x] the claims, Nos.	, as originally filed,
Nos.	, as amended under Article 19,
Nos.	, filed with the demand,
Nos. 1-16	, filed with the letter of 8.8.1997,
Nos.	, filed with the letter of,
sheets/fig	, as originally filed,, filed with the demand,, filed with the letter of, filed with the letter of
2. The amendments have resulted in the cancellation	of:
[] the description, pages	······································
[] the claims, Nos	•
[] the drawings, sheets/fig	•
3. [] This report has been established as if (some considered to go beyond the disclosure as fi	e of) the amendments had not been made, since they have been iled (Rule 70.2(c)):
4. Additional observations, if necessary:	

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

PATEMENT		
Novelty (N)	Claims 1-16	YES
	Claims	NO
Inventive Step (IS)	Claims 1-16	YES
	Claims	NO
Industrial Applicability (IA)	Claims 1-16	YES
	Claims	NO

2. CITATIONS AND EXPLANATIONS

1. The claims relate to a process for isolating a promoter functional in transgenic blackcurrant and other non-climacteric fruit, to the promoters thus isolated, to their use, to cDNA corresponding to the genes from which the promoters are derived, to the proteins encoded by the cDNA, and to vectors, cells and plants containing the promoters or cDNA. This subject-matter is novel with regard to the cited documents. It is further regarded as inventive since the available prior art does not disclose or render obvious any blackcurrant-derived promoters.

CLAIMS

- 1. A process for isolating a promoter capable of driving fruit-specific expression of DNA sequences in transgenic blackcurrant and other non-climacteric fruit comprising
- a) isolating mRNA from ripening blackcurrant fruit
- b) preparing a cDNA library from the isolated mRNA
- c) differentially screening the library from b) to identify genes expressed during the ripening period .

and

- d) screening a genomic library with probes prepared from cDNA identified according to c) to isolate the corresponding gene and its promoter region.
- 2. A promoter capable of driving fruit-specific expression of DNA sequences in transgenic blackcurrant and other non-climacteric fruit obtainable by the process of claim 1.
- 3. A promoter according to claim 2 which comprises the sequence of nucleic acid bases in Figure 9 or IDSEQ 11 (the RIB1 gene promoter) or IDSEQ 14 (the RIB 7 gene promoter)..
- 4. Promoter DNA sequences which hybridise to the DNA of claim 3 under conditions of high stringency.
- 5. cDNA for genes which exhibit differential expression in fruit during the ripening period of fruit development selected from pRIB1 (IDSEQ 1), pRIB3 (IDSEQ 3), pRIB5 (IDSEQ 5), pRIB6 (IDSEQ 7) and pRIB7 (IDSEQ 9).
- 6. DNA encoding the RIB1 or RIB 7 gene.
- 7. A vector comprising the DNA as claimed in any one of claims 2 to 6.

- 8. Use of a promoter according to claim 2,3 or 4 to control the expression of one or more genes in climacteric or non-climacteric fruit.
- 9. Use according to claim 8 wherein the non-climacteric fruit is blackcurrant.
- 10. Use of a promoter according to claim 2,3 or 4 in the transformation of plant cells.
- 11. Plant cells and plants transformed using a promoter according to claims 2,3 or 4 or a vector according to claim 7.
- 12. Plants comprising cells according to claim 11 and descendants thereof.
- 13. Plants and seeds according to claim 12 which are blackcurrants.
- 14. Use of the plants or seeds of claims 12 or 13 in the manufacture of fruit products.
- 15. A process according to claim 1 wherein the method for extracting nucleic acid from blackcurrant fruit comprises homogenising by pulping blackcurrant fruit in a buffer containing insoluble polyvinylpolypyrrolidone.
- 16. Proteins encoded by the DNA sequences of claims 5 or 6.

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(54) Title: BLACKCURRANT PROMOTERS AND GENES

(57) Abstract

Promoters and a process for isolating a promoter capable of driving fruit-specific expression of DNA sequences in transgenic blackcurrant and other non-climacteric fruit.

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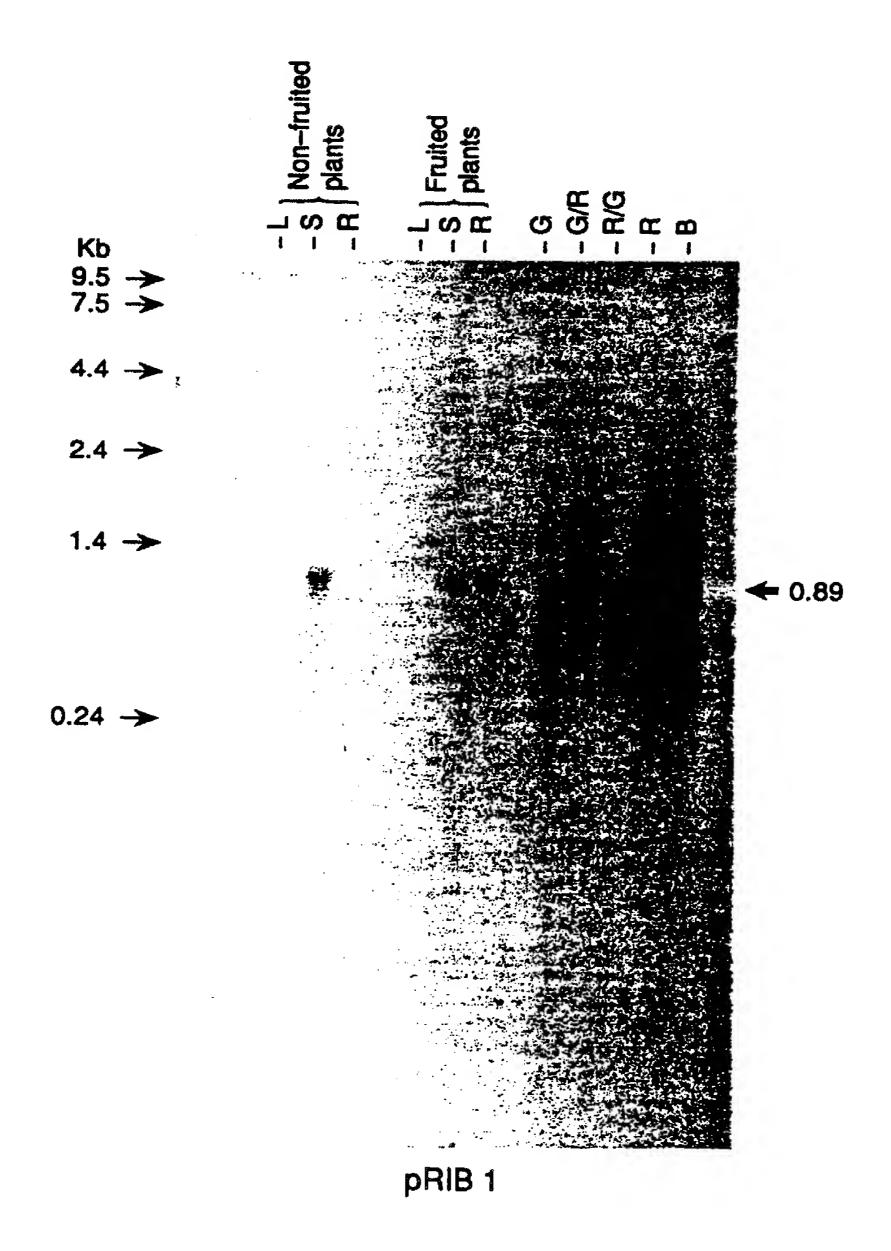


Figure 1

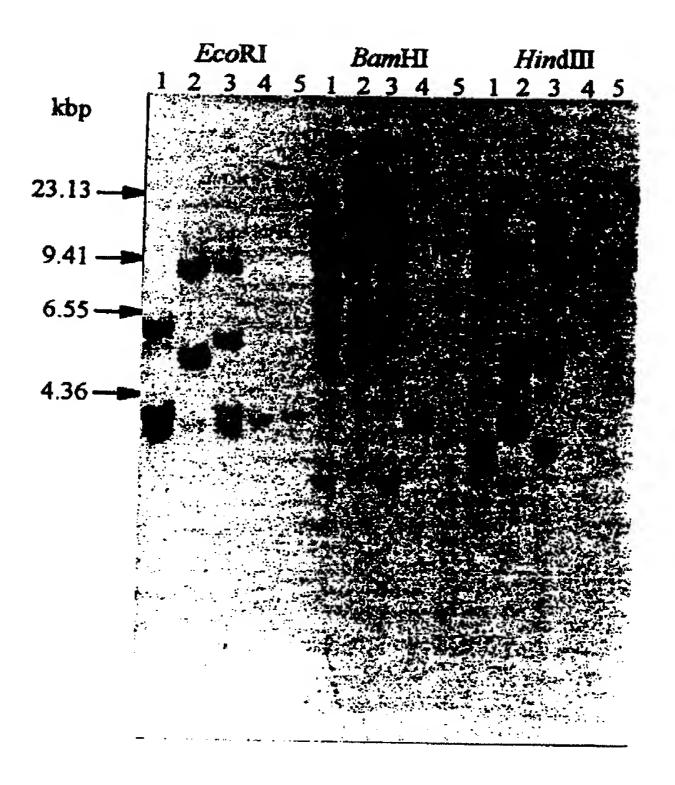


Figure 2

1 CAGCATTCCA AGAGGAAAAA AAACATGATC AAGAAGTAAT TACTACAAAA 51 GAGGAAGCTG TAGTAGTAAC TGCACCACCA CCATCAGAAA CAGCAGAGCC 101 AGCTGCAGCT GTTGTTGCCG AGGAAGAGC AACAAAGGAG CAAGAAGAGC 151 CGCCAGCAGT ATCGGCCGAG GAACCTGTGG CCCCAGCTGA AGTAGAGACA 201 AAGGTGGAAG TTACAGAAGA ACCACCAAAA GTTGAGGAGA AACCAGCAGA 251 AGTAGAGGAG GCTCCAAAGG AAACAGTAGA AACAGAACCA GCTGTTGAGA 301 AGACCATCAA GGAGGAAACT GTAGAGGACT CTGTCGTGGC ACCTGCTCCC 351 GAACCGGAAG CCGAAGTCCC AAAAGAGAAG GTAATTGCTA CTACTGAAAC 401 TACTGAGGAA GAAGAAAAG TGGCAGTTGA AGAAGTTGAA GTGAAAGTTG 451 AAACAGAGGA GGGAGAAGTT ACTGAGGAGA AGACTGAGTA AAATAAGTTG 501 TACAACTATT TTATGCACGC CTTATTTTCT CAATTGGAAG TTTATAATGT 551 AGTGGGCTTT TGGTAATATT TGGGGGTTTA ATAAGTGGTT TAAGTGGGTT 601 AAGGCTTTTT TGGAATTTAG ATATTTGGGT AAAGGCCTAC TTGAACAAAA 651 CATAGAAATT TGGCACACAT GGGTAAAAGT CAAACTTTGT TGAGGATGTT 701 TTCTTGTTGG TTAAATGTGT GTGCCAAGTA GTAGAATGTG GTGGTTGTAA 751 TGTAAGTTCT CAAGTAGGGT TTATGAGTCC TAGTATTATG CTTGATTGTA 801 TGTTGATATG AAAATGGGGG TATGTTGGCT TTGAATAAAA GTTTTTAATT 851 CC CACACAGA AAAAAAAAA AAAAAAAAT

Figure 3

- 1 AFQEEKKHDQ EVITTKEEAV VVTAPPPSET AEPAAAVVAE EETTKEQEEP
- 51 PAVSAEEPVA PAEVETKVEV TEEPPKVEEK PAEVEEAPKE TVETEPAVEK
- 101 TIKEETVEDS VVAPAPEPEA EVPKEKVIAT TETTEEEEKV AVEEVEVKVE
- 151 TEEGEVTEEK TE

Figure 4

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Figure 5

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Figure 6

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Figure 7

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Figure 8

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Figure 9

ACCATATAT TIACATIATE GEGAANCATG CTAAAAGCTF CTEGTATECA GEGAAAATGF GEFGTCAAAT CCCAAGATFC TECATGTGCC CFCTCTCTCT ACCECCATTA TEFERGOORG CECTEGIA AAAAAGAETA CAAACAAATT ACFCCTAFCA TEATTATAAA AAFAGTAGCA TAACCTCAFC TCCAATCCAC AGAGTGANTA AAGGTCATGG TCACCTACTT ACCCAACTGC ACARAACACA CAAGCACA TCCAAAAAGTA GTAGTATGAT TACACACATT TGAAAAAATG ANTACTOTICA TECCTITET TOUNDECON AFFICETICET TECANACACE TENCHANG TANANTECT TAGTANGATE ANATIFICANA TOATANCAL ATTITCTERAC AAALTAAACT ACAAGATAAT CICCCITCAG ATGATAAACT AAATGGTAGA ATATCCGTTG ACTACCCCCA ATATTIAAA ATCTCCAGCA CTAAAGAAGA GTATATCTTT TATTCATATA TCTACTTTTG ATATGACCTA AACCTTGTGT CACCCACAAT GTTGAGTACG ATGGATAATT GTTTGACTTG TOTOGOATOA GARARIGIRAT GAGACTGGCC ATTAGTTA GCCGGATGTG ATTTGGGTAT ATTGATGACA ATATAAGATA TATAAAACTT GAACAAAACA ATGUACCAAT CATACATTAT AACATAGTGT TAGGACAATA AAAGATCTTT AGTCGTAAGA GCATTAGCTC GTGACAAGAA CAAAAACGTG GATTCCCAAC THANTIGGIN INTGINATE ACTENGANC ACGININCEN TRINIGCNIC ANTGICANTG TEACAGAAN CGINACTURE GANCACATIT CGINACATGE AAGATAATAA TACCTATTAG TGTCTTTAAC ACCGGCCTAA CTFTGCATTT CTFGTCATTT GGTGACFTTT TATTGCGCAA TFGTGGCTTG AAGGAAATAA AAAGGAAAGT CTTTTTCTTG AACCCATATG GAAGCAATTT CAATGAGAGA GATAGAGAG AGGGATGGAG ATTGGGGTGG AGAATTGATA CGGATCTTCT TONGATTING AAAGTGGTTT CCACAAAAA ATATGACACA ACCCATCCAT GAACCAATAA AAACATGACA GGTCATCATT TCTTTTT TITTTCTCTC TATGACAAGG ATCTGACGTG GCAAAGAAAG AAAGTGGGTC CTGAGTCAGG TGTGTCCAT CTGTCAATAT TCTTCAAAAG AGAGTCCACC ATCTCATAGA CCANTATCAN TOTTIFICATA ANTIGOTIONE NAAGGGTGTE TITGENATIN NAAGAANGE AAGGAAAITT AGCAAGAAGT GCATTAITGG GACTGGTATA TCHARCATCA TATARGIANG GGGGAATHT ATTGGGACTC CICCAAAAQ TIATGACATT GTGATTACAC ATTIGAATGA CAGAAGTITT TGATGAAGTG GRICHTATAT TELEGRAPHE ARTHITCAAA TTACCHEATA TEFAACHCHE AACAAANTCA AGCHTITGAT CATATAARTC GAAACCAACA CACAATAATT AFGAATTECT TEGACTETET GECTETAC CAAAATACGE ACACCACAAA AAATTETTE TGFATTATAT TEGETTETA TETETAAC GETTTEGGTAT 4. Putalive promoter sequence 16.63 1401 1801 1201 1301 901 801 203 101 -=-

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1 GATCTTATATTGAGGATGCAAAGTTTCAAATTACCTGATATGTAACTCTCAACAAAATCA 60
    61 AGCTTTTGATCATAAAATCGAAACCAACACACAATAATTATGAATTTCTTTGACTCTTT 120
   121 GTCTCTGTACCAAAATACGCACACCACAAAAAATTCTTTTTGTATTATATTCGTTTTTTA 180
   241 CTCCAAAAACTTATGACATTGTGATTACACATTTGAATGACAGAAGTTTTTGATGAAGTG 300
   361 AAGGAAATTTAGCAAGAAGTGCATTATTGGGACTGGTATATATGACAAGGATCTGACGTG 420
  421 GCAAAGAAAGAAGTGGGTCCTGAGTCAGGTGTGTCCCATCTGTCAATATTCTTCAAAAG 480
  481 AGAGTCCACCATCTCATAGATGAGATTTAGAAAGTGGTTTCCACAAAAAAATATGACACA 540
  601 AAGATAATAATACCTATTAGTGTCTTTAACACCGGCCTAACTTTGCATTTCTTGTCATTT 660
  661 GGTGACTTTTATTGCCCAATTGTGGCTTGAAGGAAATAAAAAGGAAAGTCTTTTTCTTG 720
  791 AGAATTGATACGGATCTTCTTTAATTGGTATATGTAAATCACTCAGAAACACGTATACCA 840
  841 TATATGCATCAATGTCACAGAAAACGTAACTCACGAACACATTTCGTAACATGC 900
  901 ATGCACCAATCATACATTATAACATAGTGTTACGACAATAAAAGATCTTTAGTCGTAAGA 960
  961 GCATTAGCTCGTGACAAGAACAAAACGTGGATTCCCAACCTAAAGAAGGGTATATCTTT 1020
 1021 TATTCATATCTACTTTTGATATGACCTAAACCTTGTGTCACCCACAATGTTCAGTACG 1080
 1081 ATCGATAATTGTTTGACTTGTGGGGATGAGAAATGTATGAGACTGGCCATTAGTTTTA 1140
 1141 GCCGGATGTGATTTGGGTATATTGATGACAATATAAGATATAAAACTTGAACAAAACA 1200
 1201 ATTTCTCAACAAATTAAACTACAAGATAATCTCCCTTCAGATGATAAACTAAATGGTAGA 1260
 1261 ATATCCGTTGAGTACCCCCAATAATTTAAAATCTCCAGCAAATACTGTGATTCCTTTTCT 1320
 1311 TCGAAGCGAAATTCCTTCCTTCCAAACACCTTAACAAATGTAAAATTCGTTAGTAAGATT 1380
 1381 AAATTTGAAATGATAACACAAGAGTGAATAAAGGTCATGGTCACCTACTTACCCAACTGC 1440
 1441 ACAAAACACACAAGCACACCATCCAAAAGTAGTAGTATGATTACACACATTTGAAAAAAATG 1500
 1501 ACCTCCATTATTTTAGCCACCTCTCTTGTAAAAAAGATTACAAAAAAATTACTCCTATCA 1560
 1621 GCCAAACATGCTAAAAGCTTCTTGTATTCAGTGAAAATGTGGTGTCAAATCCCAAGATTC 1680
 1741 ATCAACTTGAGGGCTTTAGGACCTCTATATAAACCTCTCTCAATTGATCATCTCTGCATC 1800
1801 ACACTCTCAAGCATTCTTTCTCTCTACTTTCTTTTAGGTCAACTACACTTCCCTTTGAGT 1860
1861 TTCCAATGCCACTGTTGAGGTAAATCAAGTGATATATACATAAATTTTATTTGAAAGAT 1920
         MATVE
1921 GATTGATTCAAAGAGAACCCTTTTGTGTTTTCTTTAATAAGATCCATGTATATGAAGTTT 1980
1981 TAATGTTTCATGTTTTTTTTTTTTTTTTTTTTTTTTAATTTTGCAAT 2040
2041 ATCCCATTTGTGAAAAGATCTGTTTTCCTTTGGAAGAGATTAGAATTCGTTTCGTGTCGA 1100
2101 TTCATCATGAAAATCAATCTGGGTCTAGCTTTAATTGTGCTGATCTTGACCGGACTGTTA 2160
2161 GATGATTCGTTTTATATGTAGGCCCAATAGAGAGTGATAGTATTCCCGAAATAATACAAA 2220
2221 TCCGAGCAAACTATAATCCTCAATAGTAACTTTGTAATCTCTAAATAATCAAAAAAATAAT 2280
2281 GCTTATTGGGGTGATTGGTGTTTTGATGCAGGTTGTATCAGCGCAGACAGCATTCCAAG 2340
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2341 AGGAAAAAACATGATCAAGAAGTAATTACTACAAAAGAGGAAGCTGTAGTAGTAACTG 2400
      EKKHDQEVITTREEAVVVTA
2401 CACCACCATCAGAAACAGCAGAGCCAGCTGCAGCTGTTGTTGCCGAGGAAGAGACAA 2460
      PPPSETAEPAAAVVAEEETT
2461 CAAAGGAGCAAGAAGAGCCGCCAGCAGTATCGGCCGAGGAACCTGTGGCCCCAGCTGAAG DE20
     K E Q E E P P A V S A E E P V A P A E V
2521 TAGAGACAAAGGTGGAAGTTACAGAAGAACCACCAAAAGTTGAGGAGAAACCAGCAGAAG 2580
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2821	ATA	AG	TTG	TAC	AA	CTA:		PATO	CAC	CGCC	TTA	\TTI	TCI	CAA	TT	<b>GA</b>	4GT	TTA	TAA	TGT.	AG	2880
2881	TGG	GC	TTI	TGC	TA	ATA:	TTC	GGG	GT1	EAT?	TAA	GTG	GTI	TAA	GT(	GG:	CTA	AGG	CTT	TTT	ΓG	2940
2941	GAA	TT	TAG	AT	TT.	TGGC	TAA	AGG	CCI	'ACI	TGA	ACA	LAAA	CAT	'AG	CAA	CTT(	GGC.	AÇA(	CAT	GĢ	3000
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3061	AGA	AT(	GTG	GTC	GT.	rgta	AŢĢ	TAA	GTT	CTC	AAG	TAG	GGI	TTA	TGI	GTC	CT	AGT:	ATT	ATG(	T	3120
3121	TGA	TT(	3TA	TGI	TG.	YTAI	GAA	AAT	GGG	GGT	ATG	TTG	GCT	TTG	CAS	AAA	AG	CTT.	TTAJ	ATT:	T	3180
3181	ATA'	TA	ATA	agi	'GT <i>I</i>	TTTT	TTG	TTT	AAT	'ATC	ATT	CIT	TCA	TTC	TCI	'CGG	ATC	IAA(	CTAC	TG	T	3240
3241	CAT	CGC	CT	TGG	TA	<b>LGCT</b>	ATT	GCC	TCA	CCA	ACT.	AGC	TAA	TCG.	AAC	:GCG	AGC	CC				3292

Figure 10

# INTERNATIONAL SEARCH REPORT

inti onal Application No PCT/EP 96/04807

A. CLASS IPC 6	IFICATION OF SUBJECT MATTER C12N15/82 C12N5/10 A01H5/0	00 C07K14/415	
According	to International Patent Classification (IPC) or to both national class	sification and IPC	
<u></u>	S SEARCHED		
IPC 6	socumentation searched (classification system followed by classification s	ation symbols)	
Documenta	tion searched other than minimum documentation to the extent that	t such documents are included in the fields:	searched
Electronic	lata base consulted during the international search (name of data be	use and, where practical, search terms used)	
C. DOCUM	MENTS CONSIDERED TO BE RELEVANT		
Category *	Citation of document, with indication, where appropriate, of the	relevant passages	Relevant to claim No.
A	PLANT CELL TISSUE ORGAN CULT., vol. 24, 1991, pages 91-95, XP000618648 J. GRAHAM AND R.J. MCNICOL: "Re and transformation of Ribes" see the whole document.	generation	<b>1</b>
<b>A</b>	WO 94 21794 A (ZENECA LTD.) 29 S 1994 see pages 2-8.	eptember	1
Furt	her documents are listed in the continuation of box C.	X Patent family members are listed	in annex.
"A" docume consider filing of the citation other in the citation of the citati	ent which may throw doubts on priority claim(s) or is cited to establish the publication date of another or other special reason (as specified) ent referring to an oral disclosure, use, exhibition or	"T" later document published after the interior priority date and not in conflict we cited to understand the principle or the invention  "X" document of particular relevance; the cannot be considered novel or cannot involve an inventive step when the do "Y" document of particular relevance; the cannot be considered to involve an indocument is combined with one or ments, such combination being obvious the art.  "&" document member of the same patent	th the application but nearly underlying the claimed invention be considered to cument is taken alone claimed invention wentive step when the ore other such document to a person skilled
	March 1997	Date of mailing of the international se	arch report
Name and n	nailing address of the ISA  European Patent Office, P.B. 5818 Patentiaan 2  NL - 2280 HV Rijswijk  Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,  Fax (+31-70) 340-3016	Authorized officer Yeats, S	

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Information on patent family members

Interior No. PCT/EP 96/04807

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
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